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Characterization and Sequence of *Escherichia coli pabC*, the Gene Encoding Aminodeoxychorismate Lyase, a Pyridoxal Phosphate-Containing Enzyme

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In *Escherichia coli*, *p*-aminobenzoate (PABA) is synthesized from chorismate and glutamine in two steps. Aminodeoxychorismate synthase components I and II, encoded by *pabB* and *pabA*, respectively, convert chorismate and glutamine to 4-amino-4-deoxychorismate (ADC) and glutamate, respectively. ADC lyase, encoded by *pabC*, converts ADC to PABA and pyruvate. We reported that *pabC* had been cloned and mapped to 25 min on the *E. coli* chromosome (J. M. Green and B. P. Nichols, J. Biol. Chem. 266:12971–12975, 1991). Here we report the nucleotide sequence of *pabC*, including a portion of a sequence of a downstream open reading frame that may be cotranscribed with *pabC*. A disruption of *pabC* was constructed and transferred to the chromosome, and the *pabC* mutant strain required PABA for growth. The deduced amino acid sequence of ADC lyase is similar to those of *Bacillus subtilis* PabC and a number of amino acid transaminases. Aminodeoxychorismate lyase purified from a strain harboring an overproducing plasmid was shown to contain pyridoxal phosphate as a cofactor. This finding explains the similarity to the transaminases, which also contain pyridoxal phosphate. Expression studies revealed the size of the *pabC* gene product to be ~30 kDa, in agreement with that predicted by the nucleotide sequence data and approximately half the native molecular mass, suggesting that the native enzyme is dimeric.

In *Escherichia coli*, the aromatic pathway branch point precursor chorismate is converted to the following compounds and thereby committed to the corresponding pathways: anthranilate (tryptophan), 4-amino-4-deoxychorismate, or ADC (*p*-aminobenzoate [PABA] and folate), prephenate (tyrosine and phenylalanine), isochorismate (menaquinone and enterobactin), and *p*-hydroxybenzoate (ubiquinone).

An evolutionary relationship has been suggested for some of the chorismate-converting proteins because of similarities in the amino acid sequences. For example, the reactions that commit chorismate to anthranilate and ADC are catalyzed by two enzymes, anthranilate synthase and ADC synthase, each of which comprises two dissimilar subunits. One subunit, component I, converts chorismate and ammonia to a product, while the second subunit, component II, confers the ability to use glutamine as a source of ammonia. DNA sequence analysis revealed similarities between the two components I and the two components II, suggesting that *pabB* and *trpE* and that *pabA* and *trpG(D)* have evolved from a common ancestor (4, 12). Also, component I of each enzyme was shown to be similar to isochorismate synthase, which converts chorismate to isochorismate (20). These similarities have been interpreted to reflect not only an ancestral relationship but also presumably a relationship in the catalytic mechanisms used by these enzymes.

Unlike anthranilate, PABA is synthesized in *E. coli* from chorismate and glutamine in two steps catalyzed by two separate enzymes (19). The two subunits of ADC synthase, encoded by *pabA* and *pabB*, are required to synthesize ADC from chorismate and glutamine. ADC lyase, encoded by *pabC*, converts ADC to PABA and pyruvate (1, 6, 30, 31, 34). While in vitro data so far support a two-step reaction, it remains possible that the proteins exist in a ternary complex in vivo and that ADC synthase and ADC lyase remain active when separated. ADC lyase has been purified to homogeneity, and oligonucleotides designed from the N-terminal amino acid sequence have been used to clone and map the *pabC* gene from *E. coli* (6). While this preparation was the first homogeneous preparation of this protein from any organism, a putative *pabC* gene had been identified in *Bacillus subtilis* as part of an apparent folate operon (27). Of the three genes necessary for PABA biosynthesis in *B. subtilis*, two are highly similar to *E. coli pabA* and *pabB*.

To further our understanding of the mechanism of action of ADC lyase in *E. coli*, we have subcloned and sequenced the *pabC* gene and further characterized ADC lyase. *E. coli pabC* is similar to *B. subtilis pabC*. We show that ADC lyase contains a pyridoxal phosphate cofactor. Furthermore, a strain that is deficient in *pabC* exhibits a PABA requirement.

MATERIALS AND METHODS

Bacterial strains and plasmids. The genotypes and sources of the strains used in this work are as follows: BN102 (*trpA33* Nal^r) (18); BN117 [*his-4 proA2 argE3 rpsL704 pheA1 tyrA4 ΔtrpEA2 pabA1 pabB::Kn trpR*(Tn*10*)] (19); DH5αF′ [*supE44 ΔlacU169*(ϕ80*lacZΔM15*) *hsdR17 recA1 endA1 gyrA96 thi-1 relA1*], laboratory collection; JC7623 (*end recB21 recC22 sbcB15 thr-1 proA2 argE3 leu-6 his-4 thi-1 lacY ara-14 mtl-1 xyl-5 galK2 rpsL31 tex-33 sup-37*), laboratory collection; MG1655 (prototroph) (26); and MC1000 [Δ(*araAIOC-leu*)*7697 apaD139* Δ(*lacIPOZY-lacIPOZA*)*X74 strA galU galK*], Malcolm Casadaban. pGP1-2 was obtained from Stan Tabor (28); pMB2190, which contains a kanamycin resistance cassette, was obtained from Malcolm Casad-

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aban; pMK2004 (11) was from our laboratory collection; and pBSII SK+ was obtained from Stratagene.

Sequence analysis. The method of Henikoff (7) was used to generate nested deletions in pJMG30 (see Fig. 1). E. coli DH5αF′ was transformed with deletion plasmids, and Ap^r colonies were selected. Plasmid DNA was prepared by the boiling method (8). Plasmids were screened for insert length and for retention of the *Bss*HII sites at either end of the insert. Plasmids chosen for sequence analysis were prepared in larger quantities by the boiling method with modifications (33). Double-stranded DNA was sequenced with a Sequenase 2.0 kit, dideoxynucleotides, and [α-^{35}S]dATP in accordance with the manufacturer's instructions (United States Biochemicals, Cleveland, Ohio). Commercially available sequencing primers were generally used (United States Biochemicals); one unique synthetic oligonucleotide (5′-TT CACACAGGAAACAG-3′; Operon Technologies, Alameda, Calif.) was used to fill a gap.

Enzyme assays. Assays were performed as described previously (6, 19). The protein concentration was determined by the Bradford protein assay (2).

Nucleic acid methods. End labelling of oligonucleotides, immobilizations of lambda phage DNA, and hybridizations were done by published protocols (23). Restriction digestions were done in accordance with the recommendations of commercial suppliers. Ligations and transformations were done by published techniques (23). The heat-inducible T7 expression system of Tabor was used to express aminodeoxychorismate lyase from pBSII SK+ clones (28); labelled proteins were separated on sodium dodecyl sulfate (SDS)–12.5% polyacrylamide gels under recommended conditions (3).

pabC-specific probes were generated by end labelling of polymerase chain reaction amplification products as described previously (23). Amplification products were generated from two successive rounds of polymerase chain reaction amplification with primers deduced from the N-terminal sequence of ADC lyase. Both rounds consisted of 30 cycles with the following parameters: denaturation at 94°C for 1 min; annealing at x°C (see below) for 2 min; and extension at 72°C for 3 min. In round one, x = 55°C, the template was 1 μg of *E. coli* BN102 chromosomal DNA, and the primers were 0.77 nmol of BPN113 [5′-AA(T,C)TGNGT(G,A,T) ATNC(G,T)(G,A)TC-3′] and 0.19 nmol of BPN114 [5′-AT GTT(T,C)(T,C)TNAT(A,T,C)AA(T,C)GG-3′]. Additional reaction components were those recommended by the manufacturer for use with a Perkin-Elmer/Cetus thermal cycler. Reaction products were subsequently fractionated on 12% polyacrylamide gels, and the desired 63-bp band was eluted (23). In round two, x = 50°C, the template was approximately 0.5 ng of the eluted 63-bp band, and the primers were 0.77 nmol of BPN113 and 0.13 nmol of BPN110 [5′-AA(T,C)GGNCA(T,C)AA(G,A)CA(G,A)GA-3′]. Reaction products from round two were purified by isopropanol precipitation prior to end labelling. The final product from round two was 50 bp in length.

Determination of the pyridoxal phosphate cofactor. ADC lyase was isolated from BN117 containing pJMG30, which overproduces ADC lyase. Cells were grown for 24 h to enhance the yield of plasmid-encoded ADC lyase. The protein was purified as described previously, except that the Superose 12 column was omitted (6). The purified enzyme (0.36 mg, 1 ml), judged greater than 95% pure by polyacrylamide gel electrophoresis, was dialyzed against R buffer (50 mM Tris-HCl [pH 7.5], 5 mM $MgCl_2$, 1 mM EDTA, 10 mM 2-mercaptoethanol, 5% glycerol) and then for 10 min against

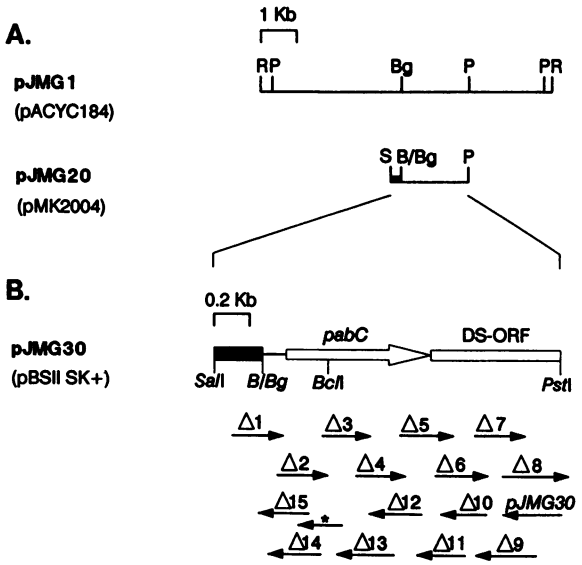

FIG. 1. Maps of clones used in this study. (A) Maps of pJMG1 and pJMG20. The solid bar represents DNA originating from pMK2004. (B) Map of pJMG30, including the sequencing strategy. The open reading frames for *pabC* and the gene encoding the double-stranded open reading frame (DS-ORF) are indicated by open bars. Deletion clones are derivatives of pJMG30; i.e., Δ1 represents pJMG30Δ1. The entire insert was sequenced on both strands. The asterisk designates a sequence obtained with a unique oligonucleotide primer. Restriction site abbreviations: R, *Eco*RI; P, *Pst*I; Bg, *Bgl*II. B/Bg in pJMG20 and pJMG30 represent a *Bam*HI site in pMK2004 that was ligated with a *Bgl*II site during the cloning of pJMG20.

R buffer containing 5 mM sodium borohydride and extensively against the same buffer lacking sodium borohydride. The spectrophotometric properties and the ADC lyase activities of the treated and untreated proteins were determined.

Cysteine reacts with pyridoxal phosphate to form an adduct with a characteristic A_{330} (24), a fact that allowed the determination of the molar ratio of the pyridoxal phosphate cofactor to the ADC lyase subunit. Purified ADC lyase (0.2 ml, 0.29 mg) or standard samples of pyridoxal phosphate (0.2 ml) were mixed with 1 ml of 200 mM cysteine (in 10 mM HCl). Samples were incubated at room temperature for 10 min, heated at 65°C for 10 min, and permitted to cool prior to the determination of the A_{330}.

Nucleotide sequence accession number. The nucleotide sequence of *pabC* has been deposited in GenBank and has been assigned accession number M93135.

RESULTS

Subcloning of *pabC*. Maps of relevant plasmids are provided in Fig. 1. *pabC* was originally isolated as pJMG1, a pACYC184 derivative containing a 7.5-kb *Eco*RI fragment obtained from lambda clone 14C1 (6, 13). To better localize *pabC* on pJMG1 and thus facilitate subcloning experiments, we used the N-terminal sequence of the protein to generate three synthetic oligonucleotides. These primers were used to amplify a 50-bp nonredundant double-stranded oligodeoxyribonucleotide that would hybridize specifically to *pabC*. The 50-bp fragment was end labelled and hybridized to a series of double digests of pJMG1 immobilized on a Zeta-Probe membrane (Bio-Rad). The sequence encoding the N

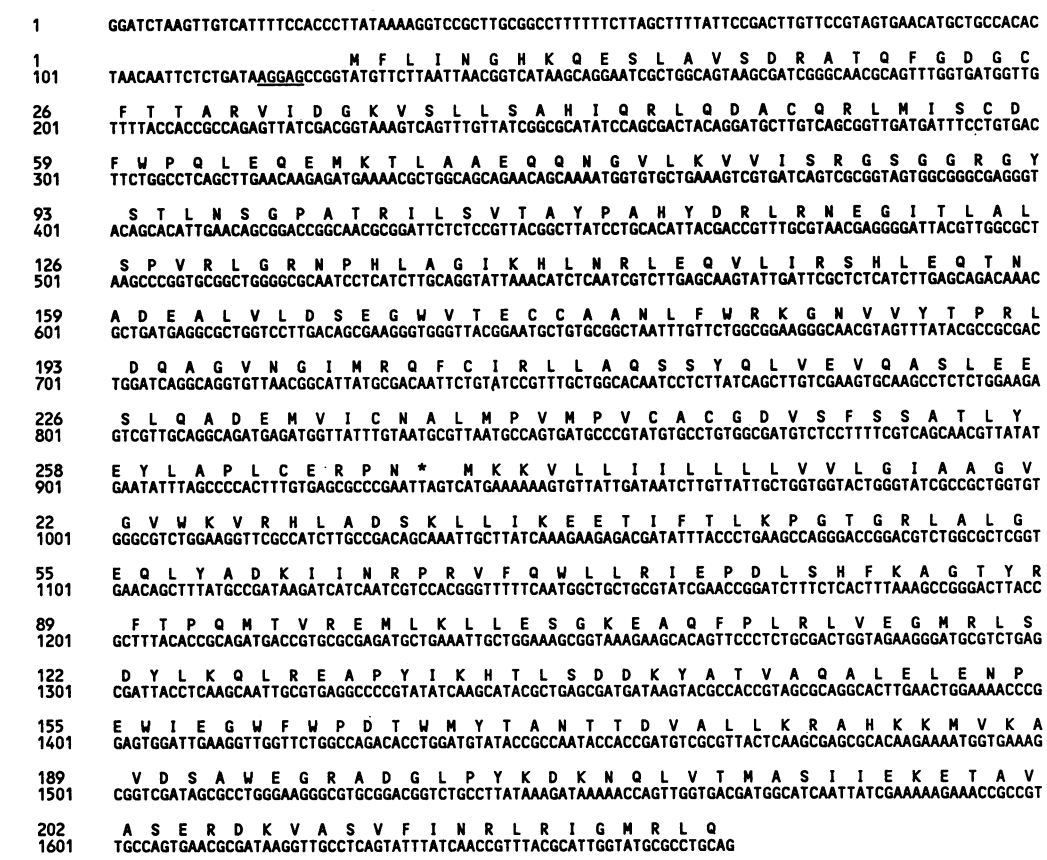

FIG. 2. Nucleotide sequence of pJMG30 (excluding the sequence originating from pMK2004). The sequence representing the putative ribosome binding site upstream of *pabC* is underlined. The amino acid sequence deduced from the nucleotide sequence is indicated by standard one-letter abbreviations, placed above the first nucleotide of the corresponding codon.

terminus of ADC lyase was thus localized to a 1.7-kb *Bgl*II-*Pst*I fragment of pJMG1 (Fig. 1).

To subclone *pabC*, we cut pJMG1 DNA with *Bgl*II and *Pst*I and ligated it with *Bam*HI-*Pst*I-restricted pMK2004. Ligation mixtures were transformed into *E. coli* DH5αF'. Twelve Kan^r Amp^s colonies were isolated, and restriction analysis revealed that 2 of these contained a 1.7-kb insert. Lysates of cells containing one of these clones, pJMG20, contained large amounts of ADC lyase, as determined by an enzyme assay.

For further work, *pabC* was subcloned into pBSII SK+. *Pst*I-*Sal*I-digested pJMG20 was ligated with *Pst*I-*Sal*I-digested pBSII K+. Following transformation into *E. coli* DH5αF', 15 Ap^r Lac$^-$ colonies were selected. Restriction analysis indicated that 11 of these contained the desired insert. Lysates of cells containing one of these clones, pJMG30, contained large amounts of ADC lyase.

Sequence analysis. Except for the fragment derived from pMK2004, the insert of clone pJMG30 was sequenced in its entirety on both DNA strands and is shown in Fig. 2. pJMG30 contained 276 bp of a pMK2004 sequence, 128 bp of a sequence upstream of *pabC*, the entire 774-bp *pabC* gene, and 728 bp of an open reading frame immediately downstream of *pabC*. The N-terminal amino acid sequence predicted by the nucleotide sequence of *pabC* agreed with that obtained by sequencing of the N terminus of the purified protein (6). The region upstream of *pabC* contains a putative ribosome binding site (AGGAG) at a position 6 to 10 bp upstream of the *pabC* initiation codon. No identifiable similarities to the consensus sequences of the −10 or −35 promotor regions of *E. coli* RNA polymerase were apparent, and without further transcription data, we could not identify unambiguously these regions. Two termination codons, spaced just one triplet apart, are possible for *pabC*. The first stop codon encountered is the relatively rare UAG. Two bases from the UAG termination codon is an apparent initiation codon for another open reading frame that extends to the end of the sequence that we have determined.

Protein expression studies. The T7 promoter lies upstream of *pabC* in pJMG30 and its derivatives. Wild-type *E. coli* (MC1000) cells were transformed with pGP1-2, which contains a heat-inducible T7 RNA polymerase gene. A Kn^r colony was selected and transformed with pJMG30 and pJMG30 deletions. pJMG30Δ9 to pJMG30Δ15 contain progressively longer deletions from the *Pst*I site towards the 3′ end of *pabC* and ultimately extending into the 3′ end of *pabC*, and these were used to define the 3′ end of the gene. Kn^r Ap^r transformants were used for expression studies. Cultures (1 ml) were grown at 32°C, induced at 42°C for 20 min, and treated with rifampin. The addition of [^{35}S]methionine resulted in the specific incorporation of the label into proteins generated from genes transcribed from the T7 promoter. Samples were analyzed by SDS-polyacrylamide gel electrophoresis. Nonradioactive lysates of the plasmid-containing strains were assayed for ADC lyase activity. The results of this experiment are shown in Fig. 3. A prominent labelled protein of about 30 kDa appeared in strains containing pJMG30, pJMG30Δ9, pJMG30Δ10, and pJMG30Δ11.

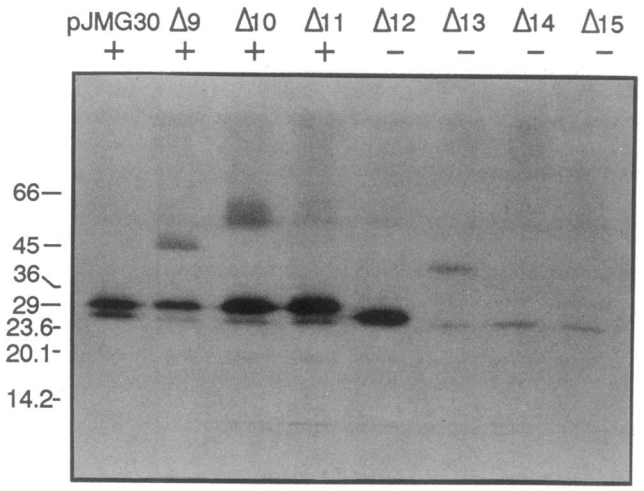

FIG. 3. Autoradiogram of a polyacrylamide gel of labelled proteins generated by cells expressing plasmids containing progressive deletions into the carboxy terminus of PabC. Expression was driven by T7 RNA polymerase as described in Materials and Methods. Molecular weight standards (in thousands) are indicated at the left. The plus and minus signs at the top of each lane indicate the presence and absence, respectively, of ADC lyase activity in cells containing the relevant clones.

Each of these plasmids also overproduced ADC lyase. A truncated protein (28 kDa) was observed in the strain containing pJMG30Δ12, and no ADC lyase activity could be demonstrated in this lysate. The endpoints of the pJMG30Δ11 and pJMG30Δ12 deletions were at nucleotides 1168 and 899, respectively, and defined the endpoint of the *pabC* open reading frame. These results are consistent with the open reading frame data shown in Fig. 2.

Physical mapping of *pabC* on the *E. coli* chromosome. *pabC* was initially cloned from a lambda clone identified by probing with a 17-bp labelled degenerate oligonucleotide designed from the N-terminal sequence of ADC lyase (6, 13). The degenerate probe hybridized strongly to lambda clones at the 25-min region of the *E. coli* chromosome and hybridized weakly to three lambda clones at 75 min (5). To investigate whether a duplicate *pabC* gene or a *pabC* homolog existed, we used pJMG30Δ12, which lacks DNA from the downstream gene, to probe a lambda clone library (13). We detected strong hybridization to two overlapping lambda clones at the known location of *pabC* (25 min), but no other region of the chromosome showed hybridization (5). We conclude that there is only one copy of *pabC* in the *E. coli* chromosome, in agreement with the phenotype of a *pabC*::*kan* strain constructed during this study (see below). The additional hybridization observed earlier may be ascribed to the degeneracy of the oligonucleotide mixture.

Construction of a *pabC* mutant. pJMG30 contained a unique *BclI* restriction site at position 371 in the coding region of *pabC* (Fig. 1). pMB2190 DNA was digested with *Bam*HI and *Eco*RI and ligated with pJMG30 DNA digested with *BclI*. Kn^r Ap^r plasmids were selected and characterized. The resultant plasmid, pUL1, was linearized with *ScaI* and used to transform *E. coli* JC7623. Kn^r Ap^s recipients were picked and tested for auxotrophy. All Kn^r Ap^s recipients required PABA in addition to the supplements required by *E. coli* JC7623. The *pabC*::*kan* allele was transferred to *E. coli* MG1655 (wild-type) cells by P1 transduction (14), and the resultant strain was designated BN1044. All Kn^r transformants required PABA for growth on minimal medium.

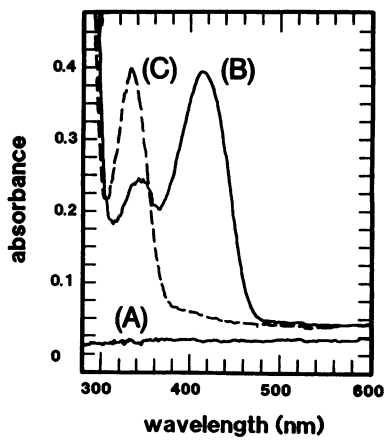

FIG. 4. Spectra of R buffer (A), native ADC lyase (B), and sodium borohydride-reduced ADC lyase (C). A Hitachi 557 double-beam spectrophotometer was used. Further details are given in Materials and Methods.

Identification of pyridoxal phosphate as a cofactor of ADC lyase. Highly purified and concentrated ADC lyase was bright yellow; the spectrum is shown in Fig. 4. Extensive dialysis of the enzyme against R buffer caused no loss of ADC lyase activity, while incubation of the purified enzyme with added pyridoxal phosphate, pyridoxal, pyridoxamine, or pyridoxamine phosphate resulted in no increase in activity (5). Reduction of the enzyme with sodium borohydride altered the spectrum and abolished ADC lyase activity (Fig. 4). The spectra of the reduced and native enzymes are similar to those of known pyridoxal phosphate-dependent proteins, including rabbit liver serine hydroxymethyltransferase (24, 25) and *E. coli* 2-amino-3-ketobutyrate coenzyme A ligase (16). We attempted to resolve native ADC lyase into the apoprotein and free cofactor by dialysis against hydroxylamine or cysteine. While some spectrophotometric changes occurred with the treatment of ADC lyase with both compounds, no more than 40% activity was lost, and the addition of pyridoxal derivatives to the treated enzyme did not restore ADC lyase activity.

Pyridoxal phosphate forms an adduct with cysteine that absorbs at 330 nm (24), and this property was used to estimate the amount of pyridoxal phosphate bound per mole of ADC lyase holoenzyme. Standard solutions of pyridoxal phosphate (0 to 600 μM) or of ADC lyase holoenzyme (0.286 mg; judged 95% pure by SDS-polyacrylamide gel electrophoresis) were mixed with cysteine as described in Materials and Methods, and the A_{330} was measured. A linear standard curve was generated from the stock pyridoxal phosphate samples, and by comparison the ADC lyase sample was determined to contain 11.4 nmol of pyridoxal phosphate. Assuming 95% purity of the initial ADC lyase sample and a subunit molecular weight of 29,700, we determined that the holoenzyme contained 1.26 mol of pyridoxal phosphate per subunit.

DISCUSSION

We report here the entire 774-bp DNA sequence of *E. coli* *pabC*, 128 bp of an upstream sequence, and 728 bp of an open reading frame immediately downstream of *pabC*. The *pabC* open reading frame encodes a protein of 29,700 dal-

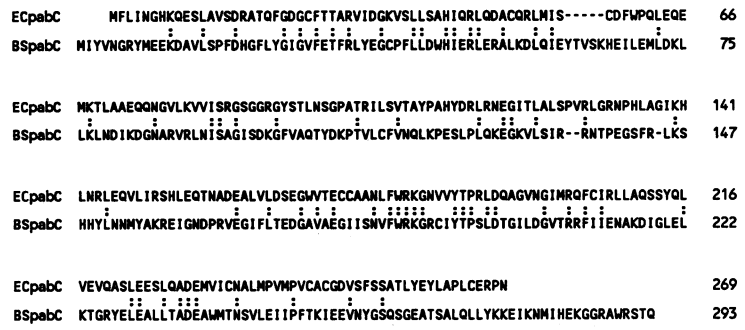

FIG. 5. Comparison of the amino acid sequences of *E. coli* PabC (ECpabC) and *B. subtilis* PabC (BSpabC). Gaps are indicated by dashes. Colons indicate amino acid identities. The sequences share 32% identity.

tons, and a protein of this mass was observed in SDS-polyacrylamide gels of labelled proteins. Since active ADC lyase elutes from gel filtration columns with an apparent molecular mass of about 50 kDa (5, 19, 34), the native enzyme is most likely dimeric. Cells harboring recombinant *pabC* plasmids overexpress *pabC*, generating aminodeoxychorismate lyase.

We found that BN1044, a wild-type strain containing a kanamycin cassette interrupting *pabC*, required only PABA to grow on minimal medium. We also probed a lambda clone library with pJMG30Δ12, which contained only *pabC* DNA (5), and detected hybridization only to the 25-min region of *E. coli*, from which *pabC* was cloned. We concluded that only one copy of the gene exists in *E. coli*. These data were surprising in that several groups, when screening for mutations in PABA biosynthesis using both chemical and transposon mutagenesis methods, only obtained mutations in the *pabA* and *pabB* genes (5, 9). The lack of detection of a *pabC* mutation may have been due to insufficient quantities of mutants having been generated and characterized or may have been due to some leakiness of a *pabC* interruption, requiring several generations of growth to deplete endogeneous PABA.

The proximity of the downstream gene to *pabC* suggests that at least these two genes may be transcribed together on a single mRNA. Since a strain containing a *pabC*::*kan* allele showed no further requirements for aerobic growth on minimal medium, the expression of the downstream gene(s) may not be essential, or this gene may be transcribed from an internal promoter.

The sequence of the downstream open reading frame was analyzed for similarity to known proteins in the GenBank data base (21). The N terminus of the deduced product of the open reading frame downstream of *pabC* exhibited 46% identity with the first 26 amino acids of *E. coli* CDP-diacylglycerol pyrophosphatase. Since this enzyme is associated with the membrane, it is possible that the downstream open reading frame product is also membrane bound (10). No other significant similarity was found.

The sequence of *pabC* was used to search for similar proteins in the GenBank data base (21). Maximum identity (32%) occurred with putative PabC of *B. subtilis* (Fig. 5). While the *B. subtilis pabC* gene product has not been shown to catalyze the ADC lyase reaction in an enzyme assay, the following characteristics support the inference: the gene exists in what appears to be a folate operon, mapping adjacent to the genes encoding ADC synthase; a strain containing the interrupted gene exhibits PABA auxotrophy; and the auxotrophy cannot be complemented with plasmids harboring the *B. subtilis* equivalent of *pabA* or *pabB* (27). The predicted product of *B. subtilis pabC* (31 kDa) is slightly larger than the *E. coli* protein (29.7 kDa).

E. coli PabC also showed 23% identity with *Bacillus sphaericus* YM-1 D-alanine aminotransferase, 23% identity with *Salmonella typhimurium* branched-chain amino acid aminotransferase, and 22% identity with *E. coli* branched-chain amino acid aminotransferase. In each case, the similarities were fairly evenly distributed throughout the sequence and were probably the result of a pyridoxal phosphate requirement of each of these proteins. Interestingly, *B. sphaericus* YM-1 D-alanine aminotransferase lysine 145, which binds pyridoxal phosphate, corresponds to lysine 140 in *E. coli* PabC (29). This region is one of high similarity between the two proteins, with 7 of 11 amino acids being identical. By analogy, lysine 140, one of six lysines in *E. coli* ADC lyase, may be involved in the binding of pyridoxal phosphate.

E. coli pabC exhibited no similarity to *trpE*, although ADC lyase catalyzes a reaction similar to a portion of the reaction catalyzed by the *trpE* gene product—the aromatization of an aminated chorismate intermediate, with the concomitant liberation of pyruvate. The intermediate analogous to ADC in anthranilate biosynthesis, 2-amino-2-deoxyisochorismate, has been synthesized independently by two groups (22, 30). Both groups showed that *Serratia marcescens* anthranilate synthase component I was kinetically competent in converting the intermediate compound to anthranilate, suggesting that this intermediate might be an enzyme-bound intermediate in the synthesis of anthranilate synthase. In addition, certain anthranilate synthase mutants accumulated aminodeoxyisochorismate, further supporting the role of this intermediate in anthranilate synthesis (15).

Unlike anthranilate synthase, ADC lyase contains a tightly bound pyridoxal phosphate cofactor. We have postulated a mechanism for the catalysis of the ADC lyase reaction in Fig. 6. Initially, ADC would bind in an imine linkage to the cofactor. An active-site base would abstract the alpha hydrogen, a process that would be stabilized by resonance with the pyridoxal phosphate. As shown, aromatization of the ring would be concomitant with release of the pyruvate. The ε-amino group of an active-site lysine could attack the PABA-pyridoxal phosphate imine to liberate PABA and restore the initial enzyme-coenzyme aldimine.

Previously proposed mechanisms for the ADC lyase reaction have not involved the use of any cofactor (32), probably because anthranilate synthase, which performs a similar reaction, does not require a cofactor. Chorismate lyase also performs a reaction very similar to the ADC lyase reaction

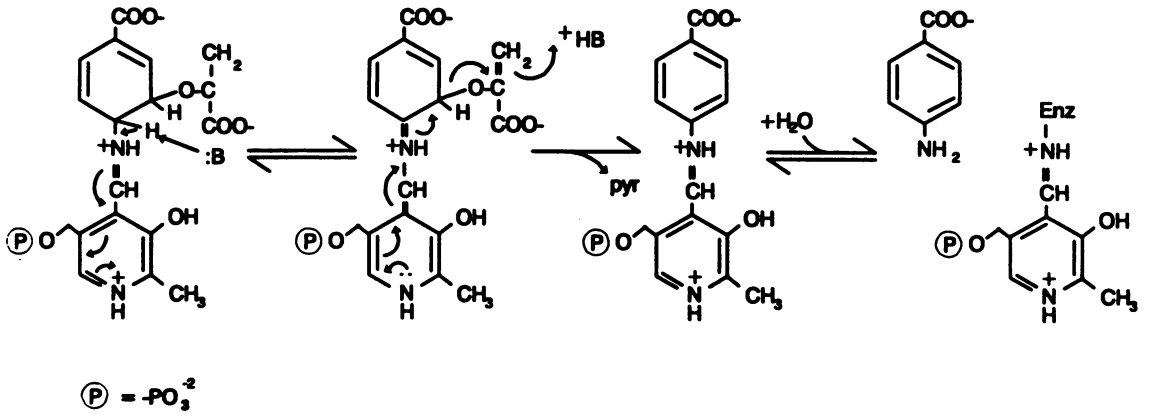

FIG. 6. Proposed mechanism for the role of pyridoxal phosphate in the ADC lyase reaction. pyr, pyruvate; Enz, enzyme.

(chorismate is converted to pyruvate and *p*-hydroxybenzoate), except that the substrate contains a hydroxyl group where ADC contains an amino group. In the accompanying paper (17), we describe the purification and characterization of chorismate lyase. We also report on the nucleotide sequence of *ubiC* and show that there is no significant similarity between ADC lyase and chorismate lyase. Therefore, ADC lyase appears to be unique among the enzymes that utilize chorismate or chorismate derivatives in requiring pyridoxal phosphate.

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